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L1: Entry 1 of 1 File: USPT Aug 17, 1993

US-PAT-NO: 5236838
DOCUMENT-IDENTIFIER: US 5236838 A

TITLE: Enzymatically active recombinant glucocerebrosidase

DATE-ISSUED: August 17, 1993

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Rasmussen; James	Boston	MA		
Barsomian; Gary	Georgetown	MA		
Bergh; Michel	Belmont	MA		

ASSIGNEE-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY	TYPE CODE
Genzyme Corporation	Cambridge	MA			02

APPL-NO: 7/ 748283
DATE FILED: August 21, 1991

PARENT-CASE:
This is a division of co-pending application Ser. No. 07/455,507, filed on Dec. 22, 1989, now abandoned, which is a continuation-in-part of application Ser. No. 289,589, filed Dec. 23, 1988, now abandoned.

INT-CL: [5] C12N 9/42, C12N 15/56, C12N 15/85
US-CL-ISSUED: 435/209; 435/69.1, 435/69.8, 435/70.3, 435/70.1, 435/172.3, 435/240.2, 435/320.1, 536/23.2, 935/14, 935/27, 935/34, 935/50, 935/70, 935/66
US-CL-CURRENT: 435/209; 435/320.1, 435/458, 435/461, 435/464, 435/466, 435/69.1, 435/69.8, 435/70.1, 435/70.3, 536/23.2
FIELD-OF-SEARCH: 435/69.1, 435/69.8, 435/70.1, 435/70.3, 435/172.3, 435/200, 435/209, 435/240.2, 435/320.1, 800/2, 800/DIG.2, 935/22, 935/23, 935/27, 935/32, 935/50, 935/70, 536/23.2

PRIOR-ART-DISCLOSED:

U.S. PATENT DOCUMENTS

Search Selected | Search ALL

	PAT-NO	ISSUE-DATE	PATENTEE-NAME	US-CL
☐	4675285	June 1987	Clark et al.	435/6
☐	4713339	December 1987	Levinson et al.	435/240.2
☐	4727138	February 1988	Goeddel et al.	536/27

OTHER PUBLICATIONS

Martin, B. et al. "Glycosylation and Processing of High Levels of Active . . . " DNA 7(2):99-106 (Mar. 1988).

Sorge, J. et al. "Complete Correction of the Enzymatic Defect of Type I . . . " Proc. Natl. Acad. Sci USA 84 pp. 906-909 (Feb. 1987).
Choudary S. et al, "The Molecular Biology of Gaucher Disease . . . " Cold Spring Harbor Symposia on Quantitative Biology vol. LI:1047-1052 (1986).
Maeda, S. "Production of Human .alpha.-Interferon in Silkworm . . . " Nature vol. 315:592-594 (Jun. 1985).

ART-UNIT: 184
PRIMARY-EXAMINER: Wax; Robert A.
ASSISTANT-EXAMINER: Prouty; Rebecca
ATTY-AGENT-FIRM: Gosz; William G.

ABSTRACT:

Recombinant enzymatically active glucocerebrosidase is produced by a eukaryotic cell. Also, a cell includes nucleic acid encoding enzymatically active glucocerebrosidase; also a eukaryotic organism contains such a cell. Also, a method for producing enzymatically active glucocerebrosidase includes steps of introducing glucocerebrosidase-encoding nucleic acid into a eukaryotic cell, causing the cell to express glucocerebrosidase, and purifying the glucocerebrosidase from the cell.

6 Claims, 12 Drawing figures

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US-CL-CURRENT: 435/209; 435/320.1, 435/458, 435/461, 435/464, 435/466, 435/69.1, 435/69.8, 435/70.1, 435/70.3, 536/23.2

CLAIMS:

We claim:
1. A method for producing enzymatically active glucocerebrosidase comprising the steps of:
introducing nucleic acid encoding human glucocerebrosidase into a CHO cell;
causing said cell to express and secrete said glucocerebrosidase into a culture medium; and
purifying said glucocerebrosidase from said culture medium.
2. The method of claim 1 wherein the pH of said culture medium is between about pH 6.5 and pH 7.2.
3. The method of claim 2 wherein the pH of said culture medium is between about pH 6.6 and pH 6.8.
4. The method of claim 1 wherein said culture medium contains O.sub.2 in an amount below about 50% saturation and sufficient to maintain the cells.
5. The method of claim 1 wherein said culture medium contains O.sub.2 in an amount between about 20% saturation and about 30% saturation.
6. A method for producing enzymatically active glucocerebrosidase comprising the steps of:
introducing a plasmid containing nucleic acid encoding glucocerebrosidase into a CHO cell, said plasmid being selected from the group consisting of pGB20, pGB37 and pGB42;
causing said cell to express and secrete said glucocerebrosidase into a culture medium; and
purifying said glucocerebrosidase from said culture medium.